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Neotypus pusillus Gregor, 1940 (Hymenoptera, Ichneumonidae) endoparasite of Maculinea nausithous (Bergsträsser, 1779) (Lepidoptera, Lycaenidae): new data on distribution in Poland with remarks on its biology

Anna M. STANKIEWICZ*, Marcin SIELEZNIEW** & Janusz SAWONIEWICZ***

*Laboratory of Social and Myrmecophilous Insects, Museum and Institute of Zoology, Polish Academy of Sciences, Wilcza 64, 00-679 Warszawa, Poland; e-mail: ams@miiz.waw.pl

**Department of Applied Entomology, SGGW – Warsaw Agriculture University, Nowoursynowska 166, 02-787 Warszawa, Poland; e-mail: sielezniew@alpha.sggw.waw.pl

***Department of Invertebrate Zoology, Institute of Biology, University of Bialystok, ul. Świerkowa 20B, 15-950 Białystok, Poland; e-mail: aptesis@uwb.edu.pl

Abstract: Neotypus pusillus in Europe is a parasite of the obligatory myrmecophilous lycaenid butterfly Maculinea nausithous. It was recorded for the last time in Poland at the beginning of the 20th century on a few sites in Lower Silesia and on one site near Poznań. During the present studies we found Neotypus pusillus in two other regions: in Polesie (eastern Poland) and in Upper Silesia. On both sites adult females were observed ovipositing on flowerheads of Sanguisorba officinalis L. with M. nausithous caterpillars inside. Additionally we reared two males from parasitized pupae. N. pusillus is probably more widespread in Poland but restricted to big populations of M. nausithous. Some remarks about the biology and ecology of the parasitoid are included in the paper.

Key words: Hymenoptera, Ichneumonidae, Neotypus pusillus, parasitoid, Maculinea nausithous, endangered species, myrmecophily

INTRODUCTION

Ichneumonidae is the biggest family of Hymenoptera, consisting of about 22,000 described species (Yu & Horstmann 1997). About 3,000 of Ichneumonidae species probably occurs in Poland and they are among poorest known insects, taking into account their biology and distribution (Sawoniewicz 2002). The larvae of almost all Ichneumonidae are parasitoids of different stages of insects with complete development, although a few species use adult spiders as hosts (Araneida) or their eggs and a few representatives may parasite Pseudoscorpionida eggs kept in a membranous sac attached to the female (Wahl 1993).

Ichneumoninae, with its 373 genera, is the second largest subfamily of Ichneumonidae and is widespread across the world (Wahl 1993). All individuals of this subfamily are endoparasitoids specializing in Lepidoptera (Hinz 1983). Two types of parasitation habits have been distinguished. Many Ichnemoninae species are idiobionts i.e. adults kill or paralyze caterpillars, which are then eaten immediately by their larvae. The second group encompasses koinobionts, which parasite active hosts, allowing them to continue development and often even to pupate. Parasitoids then reach maturity and leave the dying caterpillars or pupae. The koinobiontic genus *Neotypus* Förster, 1868 is represented in Europe by four species (Selfa & Schönitzer 1994): *N. coreensis* Uchida, 1930, *N. nobilitator* (Gravenhorst, 1807), *N. pusillus* and *N. intermedius* Mocsáry, 1883. All except the last have been recorded in Poland.

Neotypus pusillus Gregor, 1940 (syn. *Ichneumon melanocephalus* Gmelin, 1790 (preocc.) and *Neotypus melanocephalus* ssp. *pusillus* Gregor, 1940) (Selfa & Schönitzer 1994) has not been found in Poland for over seventy years. Knowledge on its distribution and ecology

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remains very fragmentary, as is the case with many other Ichneumonid species. However the interest in *N. pusillus's* life-history and behavior is still increasing due to its specific, parasitic association with *Maculinea nausithous* – an obligatorily myrmecophilous butterfly, which is endangered in Europe (van Swaay & Warren 1999). In the present studies we gathered available data about the distribution of *N. pusillus*, including information about former and newly discovered sites in Poland.

RELATION OF NEOTYPUS PUSILLUS TO MACULINEA NAUSITHOUS

Females of the genus *Neotypus* search for caterpillars which are still feeding on their host plants (Thomas & Elmes 1993). They parasitise on larvae of various Lycaenidae species (Townes *et al.* 1965, Sime & Wahl 2002). The first identification of a host of *N. pusillus* was by Torka (1928) who described it from *Lycaena arcas*, an old synonym of *Maculinea nausithous*. No other hosts have been mentioned for *N. pusillus*. Related species (Yu 1999) like *N. nobilitator* parasitizes on *Everes argiades* (Pallas, 1771), *E. lacturnus* (Godart, 1824), *Lampides boeticus* (Linnaeus, 1767), *Pseudozizeeria maha* (Kollar, 1884), *Zizina otis* (Fabricius, 1787) and for *N. coreensis* the only known host is *Niphanda fusca* (Bremer & Grey, 1852).

M. nausithous inhabits the Palaearctic region and ranges from northern Spain to the Ural Mts. and to the Caucasus (Wynhoff 1998). The butterfly is in danger of extinction in Europe and it is listed in Appendix II of the Bern Convention. Poland is the only European country where its status is estimated as 'stable' (van Swaay & Warren 1999). *M. nausithous* still occurs on numerous sites mainly in the central and southern part of the country (Buszko 1997).

Females of *M. nausithous* are on the wing in July and August and lay eggs on flowerheads of *Sanguisorba officinalis*. Young larvae feed on flowers and seeds for about three weeks. During this time they develop quickly through three instars, but they only about 1-2% of their final body weight. At the beginning of the fourth instar they drop to the ground and wait for adoption by workers of *Myrmica* Latreille, 1804 ants (Thomas 1995). Healthy as well as parasited larvae spend about 9–10 months in the ant colonies, where they prey on the ant brood. In early summer they pupate in the upper chambers of nests from which adult butterflies or parasitoids emerge after a few weeks. The relationship between *M. nausithous* and *Myrmica* ants is highly specific. In Poland, as in the whole of Europe, they survive only in *M. rubra* (Linnaeus) nests (Thomas *et al.* 1989, Stankiewicz & Sielezniew 2002).

THE OCCURRENCE OF NEOTYPUS PUSILLUS IN POLAND

N. pusillus was recorded in Poland for the first time in Lower Silesia (Duszniki Zdrój and Kudowa Zdrój) at the beginning of XIX century (Gravenhorst 1829). A hundred years later it was observed again in this region by Hedwig (1927) on sites situated in Wrocław-Leśnica, Niemcza, Kudowa Zdrój, Stronie Śląskie, Kotlina Kłodzka and in the vicinity of Strzegom. Additionally Torka (1928) found it at Prudnik and in Poznań, where he had observed an overwintering female, but in the spring he did not notice any individuals.

Specimens collected in the Białystok area and classified as *N. pusillus* (Sawoniewicz 1974) were misidentified and in fact belong to *N. coreensis* species.

NEW SITES OF NEOTYPUS PUSILLUS

During the present studies individuals of *N. pusillus* were recorded on two sites in the eastern and southern part of Poland (Fig. 1).

Kosyń (51°23'N/23°34'E, UTM FB 89, 160 m a.s.l.) is situated in Sobibór Landscape Park, Lubelska Upland, eastern Poland. The heterogeneous habitat there consists of *Molinion* and

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Filipendulion vegetation mixed with *Salicetum pentandro-cinerae*, some parts are covered by *Caricion fuscae* fens. Flowering *Sanguisorba* are scattered and their density is about 0.5 m².

In the first days of August 2001, a few females of *N. pusillus* were seen ovipositing into *S. officinalis* flowerheads. Dissections revealed the presence of 2–3 *Maculinea* larvae feeding inside each flowerhead. Apart from *M. nausithous* we found caterpillars of its close relative, *M. teleius* (Bergsträsser, 1779). There were no nests of *M. rubra* ants in the vicinity of the plants. On the site two *N. pusillus* females were collected.



Fig. 1. Past and present sites of *Neotypus pusillus* on the background of the distribution of its host Maculinea nausithous: \diamond – sites of N. pusillus from the beginning of 20th century (Hedwig 1927, Gravenhorst 1829, Torka 1928), \bigcirc – new sites of N. pusillus recorded during the present studies, \bullet – 10 km UTM squares with M. nausithous (Buszko 1997)

In early August 2002 some females of the parasitoid were observed again in another part of the site, about 0.5 km away. In July 2003 nests of *M. rubra* were searched, but from several dozens of *M. nausithous* examined, pupae only once turned out to be parasited and a male of *N. pusillus* emerged.

Głazówka near Łazy (50°24'N/19°22'E, UTM CA 88, 330 m a.s.l.) is situated on the borderland between Krakowsko-Częstochowska Upland and the Silesian Upland. The habitat encompasses *Molinion* meadows and their successional stages towards *Filipendulion* vegetation. Overgrowing is a result of abandonment of former hay meadows. *S. officinalis* is quite abundant on the site, in some parts it occurs in high density.

In July 2003 an infested pupa was found in a *M. rubra* nest, from which after 2 weeks the male of the parasitoid emerged. The same year in August a lot of females were observed laying eggs on *S. officinalis* (Fig. 2). Inside the flowerheads, similarly to Kosyń, the larvae of two sympatric species: *M. nausithous* and *M. teleius* were feeding. Furthermore a lot of parasitoid adults were recorded feeding nectar on Apiaceae flowers.



Fig. 2. A female of *Neotypus pusillus* ovipositing to a *Maculinea nausithous* larva feeding inside a *Sanguisorba* officinalis flowerhead

DISCUSSION

At present *Maculinea nausithous* is the only known host for *N. pusillus* (Yu 1999), which is regarded as highly specific (Thomas & Elmes 1993). However there is no unquestionable evidence that it is a monophagous species. Until now the parasitoid has only been recorded on a few sites in the western and northern part of the country. The new locality of *N. pusillus* in Poland found at Kosyń is also the farthest east. On the site the parasitic wasp seems to be very rare, probably as a result of the very small density of *M. rubra* nests in comparison to other *Myrmica* ants. Females of *M. nausithous* do not really have the ability to detect *M. rubra* nests (Thomas & Elmes 2001, Stankiewicz & Sielezniew 2002) and *M. nausithous* caterpillars, also those parasitized by *N. pusillus*, are adopted by unsuitable ant species. In many cases *N. pusillus* females infected *M. nausithous* larvae on plants, which grew in a completely unsuitable area lacking nests of *M. rubra*, where neither butterflies nor parasitoids could reach maturity.

At Głazówka detailed studies of ant species composition and density of their nests were not performed, but preliminary observations indicate that *M. rubra* occurs more frequently than at Kosyń, providing *M. nausithous* as well as *N. pusillus* populations with better conditions. It is also likely that *N. pusillus* is still present on some of the sites where it was discovered for the first time and in other parts of the Lower Silesian region with strong populations of *M. nausithous*. The only exception is the Poznań site where *M. nausithous* no longer occurs, and there have been no records of this species in the Wielkopolska region for the last decades (Buszko & Nowacki 2000). *N. pusillus* is probably more widespread in the southern part of the country, in Upper Silesia and in Lubelska Upland, where *M. nausithous* has a close range of distribution. However we have excluded the probability of the presence of *N. pusillus* on isolated sites near Chełmno in northern Poland, as it was not noticed during intensive behavioural studies of *M. nausithous* in August 2000 (Stankiewicz 2001).

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Sites where both M. nausithous and N. pusillus coexist are very rare since this unique relation requires specific habitat parameters, first of all density and spatial distribution of S. officinalis plants and M. rubra nests. During its life cycle the parasitoid is exposed to some limiting factors. Females of N. pusillus have to parasitize as many butterfly larvae as they can but an immature parasitoid although protected within a host has no influence on its survival. Inside a single flowerhead up to 30 eggs of N. nausithous may be found (Stankiewicz & Sielezniew 2002) because the presence of eggs does not deter females from further ovipositions into a bud. Additionally, flowerheads are often shared with eggs and larvae of M. teleius. Food competition as well as a possible cannibalistic tendency, as, for example, that exhibited by M. arion, on overcrowded plants (Thomas 1980), eliminates smaller and weaker caterpillars. Our additional observations have shown that females of N. pusillus lay eggs into S. officinalis flowerheads bearing only a few caterpillars, thereby increasing the chances of survival of the parasitoid offspring. Infected caterpillars normally develop during the endophytic phase and then their chances for adoption by Myrmica ants are probably the same as in the case of healthy Maculinea larvae. However this is a "bottleneck" in the butterfly lifecycle because of the highly specific relation with M. rubra ants, whose nests can be very sparse, as in Kosyń.

Moreover, many larvae of both *Maculinea* species can reach the same *M. rubra* nest and compete for food resources. Consequently it is possible that no individual receives enough food for effective growth and/or ant colonies abandon their nests leaving *Maculinea* inside (Thomas & Elmes 1998). There are no data on the hypothesis that infection by *N. pusillus* influences the survival of the *M. nausithous* caterpillar.

The main growth and the metamorphosis of the parasitoid occur presumably just within the pupa of the host. Adults of the parasitic wasp emerge at the same time as adults of *N. nausithous*. An interesting issue for research would be the behaviour of *N. pusillus* inside an ant colony after its eclosion from the pupa of *M. nausithous*. Ichneumon eumerus Wesm. parasiting *Maculinea rebeli* secretes special chemicals inducing mutual aggression in the ant workers. It enables safe departure from the nest for newly-emerged wasps and it also helps females of *I. eumerus* with penetration of nests during oviposition, which occurs later than in the event of *N. pusillus* (Thomas & Elmes 1993, Thomas *et al.* 2002).

Apparently not every population of *M. nausithous* can support a viable population of the parasitoid. In Europe such localities are very rare and *N. pusillus* is probably more endangered than its lepidopteran host. *M. nausithous* as well as other *Maculinea* butterflies thrive in Europe only in extensively used grassland habitats (Thomas 1995). This kind of land management is regarded as barely profitable nowadays and it is becoming rare in Poland too. Special conservation efforts are urgently needed to protect the unique butterfly – ant – parasitoid system.

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STRESZCZENIE

[Tytul: Neotypus pusillus Gregor, 1940 (Hymenoptera, Ichneumonidae) endoparazytoid modraszka nausitousa Maculinea nausithous (Bergsträsser, 1779) (Lepidoptera, Lycaenidae): nowe dane o rozmieszczeniu w Polsce wraz z informacjami o jego biologii]

Praca zawiera pierwsze od ponad 70 lat dane na temat występowania w Polsce *Neotypus pusillus* – pasożyta myrmekofilnego motyla *Maculinea nausithous* zagrożonego w Europie wyginięciem. *N. pusillus* był dotychczas wykazany jedynie z paru stanowisk na Dolnym Śląsku oraz jednego z okolic Poznania. W czasie obecnych badań znaleziono go w dwóch innych regionach: na Polesiu i na Górnym Śląsku. Na stanowiskach obserwowane były samice składające jaja do kwiatostanów *Sanguisorba officinalis* ze znajdującymi się w środku gąsienicami *M. nausithous*. Ponadto wyhodowano dwa samce z poczwarek motyla. *N. pusillus* jest prawdopodobnie bardziej rozprzestrzeniony w Polsce, ale ograniczony do du żych populacji *M. nausithous*. W pracy zawarte są również uwagi na temat biologii i ekologii tego gatu nku.

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